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The Immunizaton of White Rats Against Tularemia

Salmonella typhimurium Infection in Premature Infants

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IMMUNIZATION OF WHITE RATS AGAINST INFECTIONS WITH *PASTEURELLA TULARENSIS*[1]

By CARL L. LARSON, *Passed Assistant Surgeon United States Public Health Service*

The extreme susceptibility of most rodents and rabbits to infection with *Pasteurella tularensis* and the relative resistance of humans to infection with this agent have retarded studies of immunity in tularemia. It has not been possible, therefore, to compare the immune reactions obtained in laboratory animals with those observed in man. In order to facilitate further work efforts were made to find an animal possessing greater resistance than that of animals ordinarily employed in studying tularemia. White rats (*Rattus norvegicus*) were found to be suitable experimental animals, although their use presents some difficulties.

It is the purpose of this paper to report the results of tests comparing the susceptibility of white rats to *P. tularensis* with that of domestic rabbits, guinea pigs, and white mice, and the results obtained in white rats immunized with various vaccines and subsequently challenged with virulent strains of *P. tularensis*. Vaccines were prepared from yolk sacs of infected chicken embryos or from the products obtained by ether extraction of suspensions of infected yolk sacs or from culture.

Downs (*1*) found that domestic rabbits immunized with formalin-killed cultures of *P. tularensis* developed slight resistance to subsequent infection with this organism. Lillie and Francis (*2*) also indicated that domestic rabbits acquire only a slight degree of resistance following inoculations with attenuated strains of this organism. Many rodents and rabbits, however, possess so little natural resistance to this infection that they are not suitable for studies concerning either active or passive immunity in tularemia.

Lillie and Francis (*2*) state that the "susceptibility of white rats to highly virulent strains of *B. tularense* is low in comparison with rabbits."

[1] From the Division of Infectious Diseases, National Institute of Health.

Foshay and his co-workers (*3*) vaccinated a large number of humans with a preparation made by the oxidation of virulent strains of *P. tularensis* grown on artificial media. They report that this vaccine produced protection against tularemia, evidenced both by a decreased incidence of infection among vaccinated individuals and by amelioration of the clinical disease in those who become ill following exposure to the organism. They did not, however, produce evidence that laboratory animals were by this method protected against subsequent infection. Francis (*4*) has shown that a person recovered from tularemia is resistant to infection with *P. tularensis* and that, if a second infection does occur it is in the nature of an immune reaction such as is noted in revaccination with vaccinia virus.

MATERIALS AND METHODS

The white rats used in these studies were raised in the animal colony maintained at the National Institute of Health, Bethesda, Md. Animals of either sex weighing about 125 gm. were employed.

The strains of *P. tularensis* employed had been isolated from various sources and grown on glucose cystine blood agar. All were highly virulent for laboratory animals with the single exception of strain No. 38 which was avirulent. The virulent strains had been isolated from sputum, pleural fluid, or material from the ulcers of patients ill with tularemia. The 50-percent lethal dose of each virulent culture was determined by the method of Reed and Muench (*5*) in either white mice or white rats.

Vaccines prepared from organisms grown on artificial culture media or in the yolk sacs of chicken embryos were compared for their ability to produce immunity in rats against infections with *P. tularensis*. In order to make this comparison the following technique was employed:

Glucose cystine blood agar and chicken embryos were inoculated with identical material prepared from an actively growing culture of a virulent strain of organisms. Infected yolk sacs were obtained by a method previously described (*6*). To prepare yolk sac vaccine, the infected yolk sacs were ground in a Waring blender in sufficient 0.85-percent salt solution to make a 20-percent suspension. A loopful of this suspension was smeared on a glass slide, stained with Wayson's stain, and the approximate number of bacteria per oil immersion field was noted. To prepare culture vaccine, organisms grown on glucose cystine blood agar were added in small amounts to 0.85-percent salt solution, and their approximate numbers in the suspension also determined by examination of stained smears. Bacteria were added until about the same number of organisms were noted in stained smears of this suspension as were found in the smears of the infected yolk sac suspension. Portions from both suspensions (yolk sac and culture) were diluted with an equal quantity of salt solution. Further tenfold dilutions were made in the same diluent and the infective titre determined in mice. The remaining portion of each suspension was mixed with an equal quantity of 0.2-percent formalin or 1-percent phenol and allowed to stand for 24 hours at room temperature before being placed in the refrigerator.

Ether extraction of infected yolk sacs was performed according to the directions of Topping and Shear (7). The aqueous phase of the ether-extracted yolk sac was found to be rich in *P. tularensis*. Centrifugation of the aqueous phase in the angle centrifuge at 4,000 r. p. m. for 1 hour deposited the organisms, leaving a relatively clear supernatant fluid. The sedimented organisms were resuspended to volume in 0.1-percent formalin in salt solution. The supernatant fluid and the resuspended sediment were stored in the refrigerator for further study.

EXPERIMENTAL

White rats were studied to determine their degree of susceptibility to intraperitoneal or subcutaneous introduction of serial tenfold dilutions of suspensions containing virulent *P. tularensis*. Yolk sacs from chicken embryos infected with this organism (Mass. strain) were ground in a Waring blender and diluted decimally in 0.85-percent salt solution. Groups of two domestic rabbits and two guinea pigs each were inoculated intraperitoneally with 0.3 cc. of the varying dilution of the emulsion ranging from 10^{-6} to 10^{-12}. All rabbits receiving dilutions 10^{-6} through 10^{-10} succumbed while those receiving dilutions 10^{-11} and 10^{-12} survived. Corresponding results were obtained in guinea pigs except that one animal receiving dilution 10^{-10} survived. In addition, groups of six white rats and six white mice each were inoculated intraperitoneally with 0.3 cc. of the same material in dilutions ranging from 10^{-1} to 10^{-12}. Fifty-percent lethal end points of 5.4×10^{-7} and 3.2×10^{-10} for rats and mice, respectively, were found for this suspension of infected yolk sacs.

Similar results were obtained when a strain of *P. tularensis* (F. G.) grown on glucose cystine blood agar was employed to infect the animals. A suspension of organisms washed from the surface of slants of glucose cystine blood agar and suspended in 0.85-percent salt solution was diluted decimally in the same diluent and 0.2-cc. doses of each dilution were injected intraperitoneally into separate groups of mice, rats, and guinea pigs. The results are shown in table 1. Mice and guinea pigs were found to be highly susceptible to intraperitoneal introduction of organisms. White rats, however, had a decreased mortality rate and a prolonged survival time as compared with mice and guinea pigs.

Groups of white rats and white mice were inoculated intraperitoneally or subcutaneously with 0.3-cc. doses of serial tenfold dilutions of suspensions of virulent organisms grown on artificial media or in the yolk sac of chicken embryos. Results from two experiments given in table 2 show that white rats were less susceptible to infection than mice when tested by either route of inoculation and that rats were much less susceptible to infection when organisms were administered subcutaneously than when inoculated intraperitoneally.

TABLE 1.—*The relative susceptibility of white mice, white rats, and guinea pigs to intraperitoneal inoculation of 0.2-cc. doses of tenfold serial dilutions of a suspension of a virulent strain of* P. tularensis *(F.G.)*

Dilution of suspension	Number inoculated	Animal species tested	Number dying on certain days after inoculation										Percent dead
			2	3	4	5	6	7	9	11	13	16	
10^{-2}	6	White mice	5	1									100
10^{-4}	6	do	1	5									100
10^{-6}	6	do			5	1							100
10^{-7}	6	do			6								100
10^{-8}	6	do			4	2							100
10^{-9}	6	do				4	1						83
10^{-10}	6	do											0
10^{-2}	10	White rats	7	3									100
10^{-4}	10	do			5	4						1	100
10^{-6}	10	do			1	1	2	1		1			60
10^{-7}	10	do			1		2	2	1				60
10^{-8}	10	do					1		1	2	1		50
10^{-9}	10	do											0
10^{-10}	10	do											0
10^{-2}	2	Guinea pigs		1	1								100
10^{-4}	2	do		1		1							100
10^{-6}	2	do			1				1				100
10^{-7}	2	do			1	1							100
10^{-8}	2	do			1								50
10^{-9}	2	do			1								50
10^{-10}	2	do											0

TABLE 2.—*Titres obtained in white mice and white rats following intraperitoneal or subcutaneous inoculation with 0.3 cc. of decimal dilutions of* P. tularensis *grown on glucose cystine blood agar or in the yolk sac of chicken embryos*

Species tested	Source of organism	Strain of *P. tularensis*	Route of inoculation	50-percent lethal end point	Remarks
Mouse	Artificial media	R. H. P	I. P	3.2×10^{-7}	
Do	do	R. H. P	S. Q	5.6×10^{-7}	
Rat	do	R. H. P	I. P	7.7×10^{-3}	
Do	do	R. H. P	S. Q	0	All rats given dilutions 10^{-1} to 10^{-4} became ill but recovered.
Mouse	Yolk sac	J. J	I. P	1.8×10^{-8}	
Rat	do	J. J	I. P	3.5×10^{-7}	
Do	do	J. J	S. Q	1.9×10^{-3}	

I. P.—intraperitoneal.
S. Q.—subcutaneous.

The disease in white rats is manifested by apathy, roughness of the coat, loss of appetite, and weight loss. Frequently diarrhea and a nasal exudate are present. The weight loss is striking and may be as great as 30 percent of the original body weight. Animals developing symptoms may subsequently recover. Specific agglutination tests performed on serums obtained from recovered animals about 4 weeks after exposure to infection show the serums to have titres varying from negative to 1:640, the majority reaching 1:40 or above. As white rats do not uniformly succumb to infections with *P. tularensis*, it is necessary to employ considerable numbers of these animals in performing immunological studies.

VACCINATION EXPERIMENTS

The resistance of white rats to infection with a virulent strain of *P. tularensis* (Mass.) following intraperitoneal immunization with a living avirulent strain of organisms (No. 38) was tested. A suspension of strain No. 38 was made in 0.85 percent salt solution and adjusted to a density of T-500, a density approximating 500,000,000 organisms per cubic centimeter of suspension. Further dilutions were made in 0.85-percent salt solution and doses of 1.0 cc. of suspension containing about 500,000, 50,000,000, and 500,000,000 organisms per cubic centimeter were administered intraperitoneally to groups of 12, 10, and 9 white rats, respectively. Two weeks later these animals, together with a group of 12 untreated rats, were inoculated intraperitoneally with 0.5 cc. of a culture of virulent *P. tularensis* containing about 10,000 organisms per cubic centimeter. The results are given in table 3. They show that rats inoculated intraperitoneally with the larger

TABLE 3.—*Deaths among white rats inoculated intraperitoneally with virulent* P. tularensis (*Mass. strain*) *2 weeks following intraperitoneal injection of 2 doses of avirulent* P. tularensis (*No. 38*) *administered at weekly intervals*

Number of rats employed	Number of avirulent organisms contained in inoculum	Number of rats dying following inoculation of virulent organisms	Number of survivors yielding cultures of *P. tularensis*
12	0	12	
12	500, 000	11	0 of 1
10	50, 000, 000	2	3 of 8
9	500, 000, 000	0	4 of 9

numbers of avirulent organisms survived subsequent intraperitoneal inoculation of virulent *P. tularensis* but that rats receiving the small inocula of avirulent organisms did not survive when later infected with virulent organisms. All survivors were sacrificed 15 days after the last inoculation and the lungs, spleen, and kidneys were cultured on glucose cystine blood agar. *P. tularensis* was isolated from 7 of 17 survivors. The only lesions noted were necrotic foci in the kidneys of 3 rats and *P. tularensis* was isolated from the involved organs.

Preliminary study indicated that chemically killed vaccines prepared from yolk sacs or allantoic fluid of chicken embryos infected with a highly virulent strain of *P. tularensis* were capable of producing immunity against subsequent infection when injected intraperitoneally into white rats. Further work demonstrated that the aqueous phase resulting from ether extraction of yolk sac vaccine possessed greater immunogenic power than the crude yolk sac vaccines.

The degree of immunity attained in white rats by intraperitoneal injection of culture or yolk sac vaccines administered on 3 successive days was tested. The vaccines were made as previously described

using strain J. J. as a source of organisms. Material for titration in mice was removed from each type of vaccine prior to addition of formalin. Fifty-percent lethal end points of 1×10^{-8} and 2.2×10^{-8} per 0.3 cc. of suspension were found for the culture and yolk sac suspensions respectively. Groups of white rats were injected intraperitoneally on 3 successive days with 0.3 cc. of 10 percent normal yolk sac suspension, culture vaccine, 10 percent infected yolk sac vaccine, and the aqueous phase following ether extraction of the infected yolk sac vaccine respectively. Two weeks following the last injection of vaccine the animals were inoculated intraperitoneally with 0.3 cc. of a 10^{-3} dilution of a suspension of organisms from a culture of *P. tularensis* (strain S. A.). This suspension had a 50 percent lethal end point of 2.5×10^{-10} per 0.3 cc. for white mice. The rats were observed for 30 days before the experiment was terminated.

It was apparent that immunization with the aqueous phase of an ether-extracted infected yolk sac vaccine conferred a considerable degree of immunity on white rats under the conditions of this experiment (table 4). Immunity was manifested both by a decrease in

TABLE 4.—*Mortality in white rats tested for resistance against infection with 0.3 cc. of a 10^{-3} suspension of a virulent strain of* P. tularensis *grown on glucose cystine blood agar and administered intraperitoneally 2 weeks following the last of 3 successive daily intraperitoneal inoculations of 0.3 cc. of culture or yolk sac vaccines*

Type vaccine employed	Number of rats injected	Cumulative number of deaths in days													Total number of deaths in 30 days	Percent total mortality
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	
Normal yolk sac	29	---	---	26	---	---	---	---	---	---	---	---	---	---	---	26	89.6
Culture	29	---	---	10	13	---	15	17	18	---	19	---	---	---	20	22	75.8
Infected yolk sac	30	---	---	3	5	11	15	17	---	---	21	---	---	---	---	21	70.0
Aqueous phase of ether extracted infected yolk sac vaccine	29	---	---	1	---	---	4	7	8	---	10	---	11	13	---	13	44.8

mortality rate in animals receiving such a vaccine and by a prolongation of survival time in the animals which died as compared to the mortality rate and survival time of the control rats receiving only normal yolk sac as vaccine. The mortality rate in rats immunized with culture vaccine or infected yolk sac vaccine is high but the survival time is prolonged considerably beyond that of the controls.

In another experiment groups of white rats were given 0.3-cc. amounts of various vaccines intraperitoneally on two occasions at weekly intervals. The 10-percent infected yolk sac suspension had a 50-percent lethal end point of 3.2×10^{-8} in mice and the culture suspension had a 50-percent lethal end point of 2.4×10^{-8}. Vaccines were prepared by adding formalin to a concentration of 0.1 percent. A portion of the infected yolk sac suspension was extracted with

ether and the aqueous phase separated and retained. Groups of rats were given 10-percent normal yolk sac suspension, culture vaccine, 10-percent infected yolk sac vaccine, and aqueous phase vaccine. Two weeks after administration of the last dose of vaccines the animals were tested for immunity against a virulent strain of *P. tularensis*. A suspension of culture having a 50-percent lethal end point of 2.6×10^{-8} per 0.3 cc. in mice was prepared and 0.3 cc. of a 10^{-5} dilution of this suspension was inoculated intraperitoneally into the test animals. The results are shown in table 5. They

TABLE 5.—*Mortality in white rats tested for resistance against infection with 0.3 cc. of a 10^{-5} suspension of a virulent strain of* P. tularensis *grown on glucose cystine blood agar and administered intraperitoneally 2 weeks following the last of 2 weekly intraperitoneal inoculations of 0.3 cc. of culture or yolk sac vaccines*

Type vaccine employed	Number of rats injected	Cumulative number of deaths in days													Total number of deaths in 30 days	Percent total mortality
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	
Normal yolk sac	20	---	---	1	3	8	11	13	14	18	---	---	---	19	---	19	95
Culture	23	---	---	1	3	7	10	12	13	---	---	---	---	15	---	15	65
Infected yolk sac	23	---	---	---	---	1	---	---	---	2	---	---	---	4	---	5	22
Aqueous phase of ether extracted infected yolk sac	21	---	---	---	---	---	---	1	2	3	---	---	---	---	---	6	14

demonstrate the superiority of yolk sac vaccines over culture vaccines as here prepared in the production of immunity against tularemia in white rats.

Since vaccines derived from yolk sacs of infected chicken embryos were immunogenic the relative activity of whole yolk sac vaccine and the products resulting from ether extraction of such a vaccine were studied. A pool of infected yolk sacs which had a 50-percent lethal end point of 3.2×10^{-9} per 0.3 cc. for white mice was prepared by methods previously described. The pooled suspension of yolk sacs was divided into equal portions and to one portion sufficient formalin was added so a final concentration of 0.1 percent was attained while to the other phenol was added to a final concentration of 0.5 percent. These suspensions were allowed to stand at room temperature for 24 hours and were agitated frequently. They were then stored in the refrigerator for 4 days. At the end of this period samples to be used to immunize rats were taken from each of the suspensions. The remainder of each suspension was extracted at room temperature with twice its volume of ether and the following day the aqueous phase and the tissue phase were separated and saved. The ether was evacuated from these at room temperature and, following this, aliquot parts of the aqueous phase were removed and centrifuged at 4,000 r. p. m. for 1 hour. The supernatant fluid was retained and the sediment made up to volume in 0.1 percent formalin

or 0.5 percent phenol according to the chemical which had been used originally. Thus the following materials from yolk sac suspensions treated with formalin or phenol were made available for study: (1) 10-percent suspension of infected yolk sac, (2) aqueous phase following ether extraction of 1, (3) tissue phase following ether extraction of 1, (4) supernatant, and (5) sediment following centrifugation of 2. In addition to these the aqueous phase resulting from ether extraction of a 10-percent suspension of normal yolk sac in salt solution was available.

Separate groups of rats were inoculated intraperitoneally with 0.5 cc. each of the above materials and this procedure was repeated at the end of 7 days. Fifteen days following administration of the last dose of these materials the animals were challenged by intraperitoneal inoculation of 0.3 cc. of a 10^{-4} dilution of a suspension of a 48-hour-old culture of a virulent strain (F. R.) of *P. tularensis*. This suspension had a 50-percent lethal end point of 3.2×10^{-11} for mice and 2.7×10^{-7} for white rats when given intraperitoneally in 0.3-cc. amounts. The animals were observed for 30 days before the study was terminated. The results are given in table 6. Vaccine prepared from a

TABLE 6.—*Mortality among white rats tested for resistance against infection with 0.3 cc. of a 10^{-4} suspension of a virulent strain of* P. tularensis *grown on glucose cystine blood agar and administered intraperitoneally 15 days following the last of 2 weekly intraperitoneal inoculations of 0.5 cc. of derivatives of yolk sac vaccines killed with 0.1 percent formalin or 0.5 percent phenol*

Type of vaccine employed	Killing agent employed	Number of rats injected	Cumulative number of deaths in days													Total number of deaths in 30 days	Percent total mortality
			1	2	3	4	5	6	7	8	9	10	11	12	13	14	
Normal yolk sac	Formalin (0.1 percent).	30	--	18	--	22	24	26	--	--	--	27	--	28	--	--	28	93.3
Infected yolk sac	do	29	--	2	3	4	8	11	14	15	--	16	--	--	17	18	19	65.5
Tissue phase [1]	do	27	--	--	1	7	10	11	17	--	--	19	--	--	--	20	20	74.1
Aqueous phase [1]	do	29	--	--	--	--	--	--	1	--	--	7	8	--	--	9	9	31.0
Sediment of aqueous phase.[1]	do	29	--	--	1	2	5	--	6	9	--	10	11	--	--	--	12	41.4
Supernatant of aqueous phase.[1]	do	29	--	--	--	2	3	5	7	--	--	8	9	--	--	10	11	37.9
Infected yolk sac	Phenol (0.5 percent).	28	1	2	6	10	12	14	15	--	--	--	--	--	--	--	15	53.6
Tissue phase [1]	do	27	--	--	3	6	8	12	14	19	--	21	--	--	22	--	22	81.5
Aqueous phase [1]	do	28	--	--	--	--	1	2	6	7	--	13	--	--	--	--	13	46.4
Sediment of aqueous phase.[1]	do	28	--	--	1	3	4	5	8	9	--	--	10	11	--	12	12	42.9
Supernatant of aqueous phase.[1]	do	27	--	--	--	1	3	6	--	--	--	--	--	--	--	--	6	22.2

[1] Antigen derived by ether extraction of 10 percent infected yolk sac suspension vaccine.

10-percent suspension of infected yolk sac possessed slight immunizing ability. The aqueous phase obtained by ether extraction of the crude yolk sac vaccine had marked immunizing ability. The sediment resulting from centrifugation of the aqueous phase for 1 hour at 4,000 r. p. m. in the angle centrifuge contained many organisms and the supernatant contained negligible numbers of bacteria. Both the former and latter were approximately as active as the whole aqueous phase when employed to produce active immunity in rats.

SOLUBLE ANTIGEN

It appeared that a soluble substance was associated with *P. tularensis* and that it was capable of stimulating the production of protective antibodies against tularemic infections in white rats. Complement fixation tests were performed on some of the above preparations, employing the technique described by Bengtson (*8*). The serum was one obtained from an individual recently recovered from tularemia and had an agglutination titre of 1:1,280 against strain No. 38 of *P. tularensis*. The antigens used were the aqueous phase, the sediment of the aqueous phase, and the supernatant from the aqueous phase of both the formalin- and phenol-treated 10-percent infected yolk sac suspensions used in the immunization of the rats previously discussed. The antigens were titred against a 1:8 dilution of serum. It was found (table 7) that an antigen capable of entering into a comple-

TABLE 7.—*The titre of complement-fixing antigen in the fractions of ether-extracted 10 percent infected yolk sac suspensions*

Antigen	Chemical treatment	Titre of antigen
Aqueous phase	Formalin	1:128
Sediment of aqueous phase	do	1:8
Supernatant of aqueous phase	do	1:128
Aqueous phase	Phenol	1:128
Sediment of aqueous phase	do	1:2
Supernatant of aqueous phase	do	1:128

ment fixation reaction was associated with the aqueous phase following ether extraction of infected yolk sac suspensions and that this antigen could be separated in large part from the organisms by centrifugation. There was little of this antigen present in the sediment but a large amount was present in the supernatant fluid.

DISCUSSION

Studies of the susceptibility of white rats to infections with *P. tularensis* show these animals to possess such a degree of resistance to infections with this agent that it is possible to perform studies concerning the immune reactions produced when rats are infected with this organism. Vaccines have been produced by harvesting the yolk sacs of infected chicken embryos, killing the organisms by chemical means, and ether extracting the yolk sac suspensions. The aqueous phase obtained by this technique is very active immunogenically and tends to prevent fatal infections in white rats due to subsequent inoculation of virulent strains of *P. tularensis*. None of the other antigens which have been studied in this laboratory have approached the effectiveness of aqueous-phase vaccines as immunizing agents. A soluble substance is present in the aqueous phase of these prepara-

tions; it can be freed of suspended bacteria by centrifugation of the aqueous phase. White rats immunized with the soluble substance are markedly resistant to infections with *P. tularensis*. The soluble substance is capable of acting as a specific antigen in complement fixation tests using human convalescent serum as antibody.

SUMMARY

The resistance of white rats to infection with *P. tularensis* was found to be of such a magnitude that they could be employed as test animals in studying immunity in tularemia.

Active immunity to infections with *P. tularensis* was produced in white rats by the administration of vaccines prepared from yolk sacs of infected chicken embryos.

Ether extraction of infected yolk sac suspensions enhanced the antigenic value of yolk sac vaccines.

A specific soluble antigen was demonstrated in the aqueous phase of ether-extracted yolk sac vaccines.

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STUDIES OF THE ACUTE DIARRHEAL DISEASES

XVI. AN OUTBREAK OF *SALMONELLA TYPHIMURIUM* INFECTION AMONG NEWBORN PREMATURE INFANTS [1]

By JAMES WATT, *Surgeon*, and ELIZABETH CARLTON, *Junior Bacteriologist*, *United States Public Health Service*

There has been a wide interest in recent years in outbreaks of diarrheal disease among the newborn. The epidemics described have been predominantly of unknown etiology. Neither the source of the infection nor the modes of spread could be ascertained. This small

[1] From the Division of Infectious Diseases, National Institute of Health, and the Charity Hospital of Louisiana at New Orleans.

outbreak, six infections, is unusual in that these factors were all clearly evident.

There is only one reference in the literature of the United States to an outbreak of this type occurring in the first month of infancy. A previously undescribed member of the genus *Salmonella* (*S. wichita*) was isolated from an epidemic of the newborn in Wichita, Kans., by McKinlay and was identified by Schiff and Strauss (*1*, *2*). The method of introduction of this infection into the nursery was not ascertained in this epidemic.

EPIDEMIOLOGY

On November 6, 1943, a colored female was admitted to the obstetrical service of the New Orleans Charity Hospital. Delivery of a premature infant was uneventful. Six hours after delivery the mother's temperature rose to 105° and a profuse watery diarrhea began. Nausea, vomiting, general malaise, and anorexia were marked. She reported at this time that she had had a moderate diarrhea for 3 days before admission. Fecal cultures obtained on November 8 were found to be positive for *Salmonella typhimurium*.[2] Her temperature returned to normal within 24 hours but the enteric disorder persisted for 11 days. Stool cultures were repeatedly positive.

The premature infant, following delivery, was removed at once to the nursery and subsequently had no contact whatsoever with his mother. No abnormality in the infant was noted for the first 3 days. The child then passed a watery green stool with mucus and in the following 24 hours had two additional bowel movements of similar type. Death, which occurred suddenly on November 10 prior to bacteriological diagnosis of the mother's illness, was at first attributed to an aspiration pneumonia. The stool abnormality was not reported to the laboratory and was noted during the investigation of the ensuing outbreak. Also at a later date it was ascertained that the body of the child had remained unclaimed in the morgue and permission for post-mortem study was obtained on December 2, more than 3 weeks after death. No gross anatomical lesions were found to explain the death. The lungs were clear. There were pronounced post-mortem changes in the intestines, which would have obscured other possible pathological lesions. Cultures were taken from the contents of the heart and from the intestines. *S. typhimurium* was isolated from both.

The first reported case of diarrhea in the premature nursery was on November 14, 4 days following the death of the preceding case. A second mild case was reported on the following day. These children were cultured routinely and the first test, performed without enrichment medium, was negative. Subsequently both were found positive

[2] The final identification of the *Salmonella* strains was made by Dr. P. R. Edwards, Agricultural Experiment Station, University of Kentucky, Lexington, Ky.

for *S. typhimurium*. A more intensive study of the outbreak was begun after a third child had become critically ill with fever and diarrhea. Cultures of the blood and stool of this child, obtained on November 18, were both found to be positive for *S. typhimurium*. All infants in the ward were then examined culturally on November 22. Four, the three above and one other (P2), were culturally positive for *S. typhimurium* at that time and repeatedly thereafter. These five infected infants (including the original case which died) had been cared for in five adjacent bassinets, two on each side of the initial fatal case. The responsibility for the care of the infants in the nursery is divided between the services of two medical schools and an independent service. The five infected infants were on the same service.

One additional infection, the sixth, was identified. An infant, discharged from the same nursery on November 12, was readmitted on November 24 with a history of diarrhea since discharge. The bassinet used for this child was about 15 feet from the original case in the same large unpartitioned ward. Medical care for this baby was provided by a different service but nursing care was the same for all.

The source of infection in the mother of the initial case (P1) was not determined. Her child presumably acquired the infection at birth since there was no subsequent contact between the mother and child. A spread of infection in the ward evidently accounted for the remaining cases. The general plan of operation in the nursery was such that only one of the three routes by which enteric infections are usually spread could be suspected. Food, including water, was prepared in individual bottles in a central kitchen for the entire pediatric service of which this nursery formed only a small part. The nipples were sterilized in individual cellophane packets by autoclaving. Flies were not present. The location of the five cases in adjacent cribs provides support for the belief that the infection was transported by the fingers of attendants who provided care to the different infants one after another. Bacteriological evidence was also obtained which indicated further the probability of transmission of enteric organisms by fingers. Cultures of the fingers of five of the nursery workers who were going about their usual duties (not just after changing diapers) were positive for *Escherichia coli*. It appears, therefore, that this specific enteric infection was acquired from the mother at birth and was introduced to the nursery by the infected infant. In the nursery the infection was spread from this baby to infants in four adjacent bassinets and to one other in a bassinet nearby, presumably by the contaminated fingers of those caring for the babies.

BACTERIOLOGICAL DIAGNOSIS

The practical importance of two diagnostic cultural procedures was emphasized in the study of this epidemic. (1) Tetrathionate

enrichment broth was found to have particular value in the isolation of *S. typhimurium* from the infants. The cultures were taken by a rectal swab which was used to streak an S. S. agar plate. This same swab was then placed in 3 cc. of tetrathionate broth. A comparison of the two methods in 77 cultures on positive infants showed 29 positive by direct plating and 56 positive by enrichment. (2) S. S. agar was less inhibitive for the nonpathogenic organisms in the stools of these milk-fed infants than for those of older children and adults. Also, the lactobacilli frequently caused a clouding of the agar, making it difficult to detect the colonies of possible pathogens. It is important, therefore, in the examination of infants' stools to use an enrichment medium such as tetrathionate broth, and to use a much smaller inoculum in direct plating than should be used for older children and adults. Satisfactory plates can usually be obtained by inoculating about one-fourth of the plate with the original swab. A sterile swab is then used to spread the inoculum over the remainder of the plate and also to a second plate.

Positive blood cultures were obtained on two of these cases (P1 and P5). The heart's blood obtained at post-mortem examination of P1 gave a pure culture of *S. typhimurium* on both brain broth and tetrathionate broth. The failure to find any other organisms by this technique is evidence that the bacteremia was ante mortem rather than post mortem. Blood cultures were not taken on the other cases.

Three of the five infants were followed bacteriologically until culturally negative; the last positive tests were obtained on December 3, 17, and 29. Two infants and the mother of the first case were discharged on December 6, January 26, and December 27, respectively, while still positive. One infant (P1) died on November 10 when culturally positive but these cultures were not obtained until 21 days after death.

CLINICAL

The temperature records of the five babies followed throughout their clinical illiness in the premature nursery are shown in figure 1, together with the positive cultures found and the days of recorded diarrhea. Their ages at the time of first potential exposure are given. One death occurred (P1), two others (P3 and P5) recovered clinically and bacteriologically, and two were discharged as convalescent carriers (P2 and P4).

The character and severity of the illness varied widely. The mildness of symtoms even in premature infants was notable in three cases. One child (P2) had no symtoms other than moderate fever, which was considered insignificant, until the positive stool culture had been obtained. The child nursed normally and continued to gain weight, and apart from the slight elevation of temperature had no indication of illness. In another child (P3) there was, in addition to fever, a

moderate diarrhea with stools which were greenish and contained mucus. The diarrhea persisted for 2 weeks, in the first week with five to seven mucoid greenish stools daily, and in the second gradually returning to normal. This child also did not appear ill at any time and gained steadily throughout the duration of the infection. The history of another child (P4) was similar, though a mild diarrhea con-

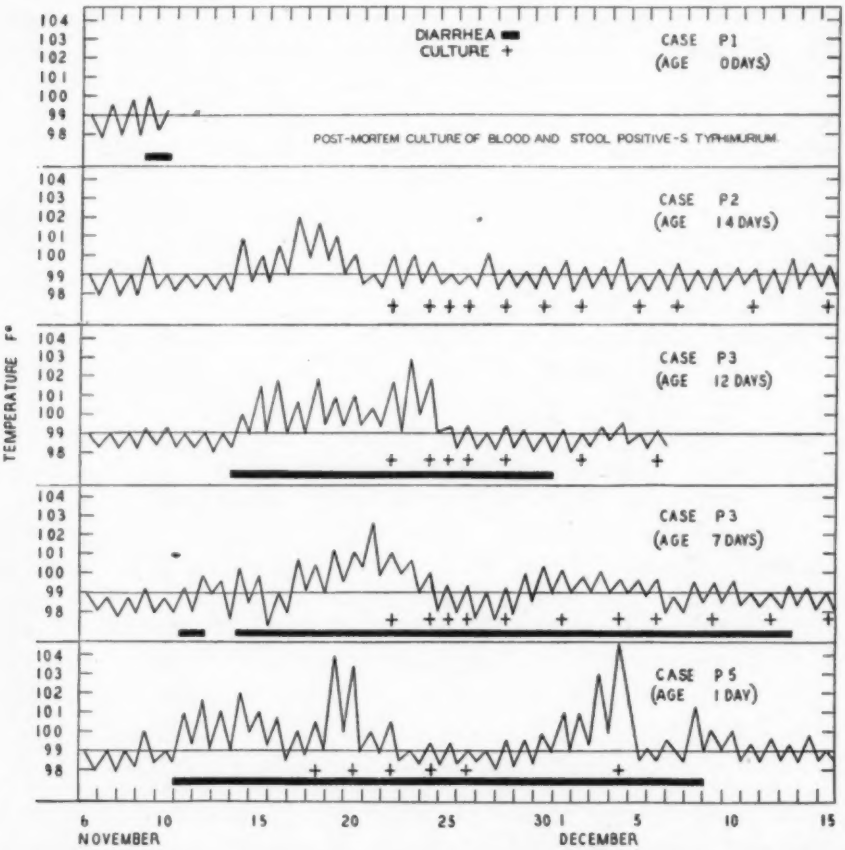

FIGURE 1.—Temperature records of 5 cases, showing age at time of first potential exposure, days of recorded diarrhea, and positive cultures of *S. typhimurium*. One death occurred (P1), 2 recovered clinically and bacteriologically (P3 and P5), and 2 were discharged as convalescent carriers (P2 and P4).

tinued for 4 weeks and at onset some blood was noticed in the stools. The child was listless during the 3 days of maximum temperature elevation but otherwise did not appear ill.

In contrast with these mild cases, P5 became critically ill. Diarrhea began on November 11 and progressively increased in severity. Mucus and blood were observed in the stools. At the end of 1 week, the child was obviously ill, listless, and moderately dehydrated. In this child hypodermoclyses, transfusion, and an oxygen tent were all

required. Sulfadiazine was given. Improvement was marked by the end of the second week of illness. There was, however, a recurrence of fever, but there were no severe general manifestations. The stools in this child were abnormal for almost 1 month—from November 10 to December 8. The organisms were isolated from two blood cultures taken on November 18 and 20.

P6, the child ill at home and for that reason not included in the chart, was reported to have had a persisting diarrhea and a little fever for 2 weeks. On admission to the hospital this child was moderately ill but the diarrhea slowly improved and fecal cultures became negative.

The mother had an acute diarrheal disorder with pronounced elevation of temperature shortly after delivery. The diarrhea, quite mild for 3 days before delivery, was very severe for the next 4 days. Gradual improvement was then evident and clinical recovery was complete by the twelfth post-partum day.

CONTROL MEASURES

Reliance was placed upon a combination of bacteriological and general control measures. All infants in the nursery were cultured for the identification of missed cases and carriers. One of the infections (P2) would have remained undetected except for this procedure. Infants with even one possibly abnormal loose stool were cultured immediately. The known infected infants were placed under strict isolation in a separate nursery. The general measures of control were directed primarily to an improvement of hand washing practices. Washing of hands was required, without exception, following the handling of each infant.

COMMENT

The possibility of introduction of enteric pathogens into nurseries by the infection of infants at birth is not considered, as a general rule. Further evidence that this does occur is shown by a *Shigella* infection observed in the same nursery. A mother who had given birth to a premature infant developed diarrhea and was culturally positive for *Shigella sonnei*. Her infant, after birth, was immediately sent to the premature ward and had no further contact with the mother. This child developed diarrhea on the third day after delivery and his cultures were also positive for *S. sonnei*. There was no detected spread within the nursery.

SUMMARY

A small outbreak of *S. typhimurium* infection in a premature nursery is described. The infection was introduced by a child, presumably infected at birth, and was then spread to four children in adjacent

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bassinets and to one in a nearby bassinet. Cases were identified by use of rectal swabs plating directly to S. S. agar and to tetrathionate enrichment broth. Clinical manifestations varied from a rapidly fatal illness to mild, almost asymptomatic, infections. Control measures consisted of repeated cultural surveys of the whole ward, isolation of all infants found positive, and such general measures as effective hand washing after handling any child.

ACKNOWLEDGMENT

The authors wish to acknowledge their indebtedness to Dr. O. P. Daly, Director, for permission to conduct this and other studies in the Charity Hospital at New Orleans, to Dr. Emma S. Moss, Director of Pathology, for the use of facilities in her department, and to the chiefs of service and other physicians for their active cooperation in the furtherance of these investigations.

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DEATHS DURING WEEK ENDED JUNE 2, 1945

[From the Weekly Mortality Index, issued by the Bureau of the Census, Department of Commerce]

	Week ended June 2, 1945	Corresponding week, 1944
Data for 92 large cities of the United States:		
Total deaths	8,231	8,005
Average for 3 prior years	8,138	
Total deaths, first 22 weeks of year	196,515	201,762
Deaths under 1 year of age	512	569
Average for 3 prior years	583	
Deaths under 1 year of age, first 22 weeks of year	12,964	13,052
Data from industrial insurance companies:		
Policies in force	67,350,674	66,588,800
Number of death claims	11,737	10,648
Death claims per 1,000 policies in force, annual rate	9.1	8.4
Death claims per 1,000 policies, first 22 weeks of year, annual rate	10.9	10.8

PREVALENCE OF DISEASE

No health department, State or local, can effectively prevent or control disease without knowledge of when, where, and under what conditions cases are occurring

UNITED STATES

REPORTS FROM STATES FOR WEEK ENDED JUNE 9, 1945

Summary

Of the total of 92 cases of poliomyelitis reported for the current week, as compared with 71 last week and 41 for the 5-year (1940–44) median, 42 cases occurred in Texas (last week 24), 13 in California (last week 3), and 11 in New York (last week 3). The other 26 cases occurred in 18 States, only one of which (South Carolina) reported as many as 3 cases.

Of the total of 903 cases reported to date this year, as compared with 586 for the same period last year, which latter number was also the 5-year median for the period, an aggregate of 398 cases occurred in 3 States (last year's corresponding figures in parentheses), as follows: New York 165 (39), Texas 161 (63), California 72 (118). For the same period last year Louisiana and Washington had reported totals of 47 and 33 cases, respectively. The cumulative total for the 12-week period since March 17, the week of lowest incidence this year to date, is 505, as compared with 323 for the corresponding period last year.

A total of 143 cases of meningococcus meningitis was reported, as compared with 171 last week, 314 for the corresponding week last year, and 75 for the 5-year median. Only 3 States reported more than 10 cases each—New York (21), Ohio (14), and Pennsylvania (13). Decreases were recorded in 7 of the 9 geographic divisions. A slight increase (33 to 37) occurred in the East North Central division, and the same number (13) as for last week was reported in the Pacific area.

A total of 90 cases of Rocky Mountain spotted fever has been reported for the year to date, as compared with 89 for the same period last year and a 5-year median of 127. Figures by geographic divisions for the year are as follows (last year's corresponding figures in parentheses): Middle Atlantic 10 (12), East North Central 9 (1), West North Central 2 (4), South Atlantic 41 (36), East South Central 6 (2), West South Central 1 (4), Mountain 18 (29), Pacific 3 (1). The incidence of tularemia to date is about 41 percent above that for last year—363 cases as compared with 257 last year. The incidence of both smallpox and typhoid fever is below that for last year.

A total of 8,890 deaths was recorded in 93 large cities of the United States, as compared with 8,680 last week, 8,588 for the 3-year (1942–44) average, and 8,360 for the corresponding week last year. The cumulative total is 216,604, as compared with 222,122 for the corresponding period last year.

Telegraphic morbidity reports from State health officers for the week ended June 9, 1945, and comparison with corresponding week of 1944 and 5-year median

In these tables a zero indicates a definite report, while leaders imply that, although none was reported, cases may have occurred.

Division and State	Diphtheria			Influenza			Measles			Meningitis, meningococcus		
	Week ended—		Median 1940–44	Week ended—		Median 1940–44	Week ended—		Median 1940–44	Week ended—		Median 1940–44
	June 9, 1945	June 10, 1944		June 9, 1945	June 10, 1944		June 9, 1945	June 10, 1944		June 9, 1945	June 10, 1944	
NEW ENGLAND												
Maine	0	0	0	------	------	------	3	139	139	1	1	1
New Hampshire	0	0	0	------	------	------	3	3	5	0	0	0
Vermont	5	0	0	------	------	------	31	14	85	0	0	0
Massachusetts	4	1	2	------	------	------	354	877	1,078	7	13	3
Rhode Island	0	0	0	19	16	------	11	6	81	1	2	0
Connecticut	1	1	0	1	------	------	89	425	342	1	1	1
MIDDLE ATLANTIC												
New York	10	6	8	[1] 2	(1)	[1] 3	142	1,053	1,268	21	36	19
New Jersey	4	5	5	2	1	3	57	713	1,256	3	10	2
Pennsylvania	11	10	14	1	4	------	620	411	715	13	26	5
EAST NORTH CENTRAL												
Ohio	4	2	7	4	2	3	53	162	315	14	36	2
Indiana	1	5	4	13	1	1	49	51	73	0	4	2
Illinois	4	1	20	7	9	9	401	345	345	10	19	1
Michigan [2]	9	6	4	------	------	1	251	447	832	5	21	2
Wisconsin	0	3	3	21	6	16	155	1,431	1,431	5	9	1
WEST NORTH CENTRAL												
Minnesota	1	2	1	------	------	------	17	324	309	1	3	0
Iowa	4	2	3	------	------	------	63	105	105	1	0	0
Missouri	3	0	0	11	------	------	45	62	185	6	9	3
North Dakota	1	3	1	1	------	2	2	21	21	0	0	0
South Dakota	4	3	1	------	------	------	36	5	7	1	0	0
Nebraska	2	2	1	------	------	1	105	29	29	1	0	0
Kansas	2	3	3	------	------	1	41	133	287	1	3	1
SOUTH ATLANTIC												
Delaware	0	0	0	------	------	------	4	10	10	0	1	0
Maryland [2]	12	6	4	1	1	1	25	204	204	0	8	8
District of Columbia	0	1	1	------	------	------	2	60	60	1	2	1
Virginia	3	3	3	76	43	60	32	280	280	3	3	1
West Virginia	1	1	3	------	7	4	6	250	33	2	2	2
North Carolina	2	4	5	------	8	2	29	327	262	2	3	2
South Carolina	7	3	3	74	109	89	32	188	77	1	1	1
Georgia	4	3	3	5	------	8	4	37	97	4	2	0
Florida	1	2	2	------	1	2	1	111	71	1	7	0
EAST SOUTH CENTRAL												
Kentucky	0	1	2	------	------	2	13	42	63	1	9	1
Tennessee	0	2	2	23	10	11	63	72	103	6	10	2
Alabama	4	2	2	9	9	14	5	71	71	5	0	1
Mississippi [2]	5	4	3	------	------	------	------	------	------	1	9	0
WEST SOUTH CENTRAL												
Arkansas	2	4	4	15	12	12	61	163	68	3	1	0
Louisiana	0	8	3	------	4	2	52	21	18	0	6	2
Oklahoma	9	0	2	31	47	10	33	180	38	2	3	0
Texas	28	22	20	393	287	245	271	1,172	516	3	20	3
MOUNTAIN												
Montana	0	0	0	3	4	1	7	43	86	0	0	0
Idaho	0	0	0	3	2	------	2	11	29	0	0	0
Wyoming	0	0	0	------	------	1	12	48	15	1	0	0
Colorado	7	8	8	63	12	18	10	103	151	0	2	0
New Mexico	2	1	1	------	------	1	1	58	38	0	0	0
Arizona	1	0	2	33	33	45	11	112	64	1	1	0
Utah [2]	0	0	0	------	1	------	212	71	112	1	1	0
Nevada	0	0	0	------	------	------	4	4	13	0	0	0
PACIFIC												
Washington	4	1	1	------	2	2	193	223	263	4	2	1
Oregon	5	0	1	7	3	7	89	111	105	0	0	2
California	11	23	16	13	42	49	1,458	3,384	1,163	9	28	4
Total	178	154	192	831	676	731	5,160	14,112	14,662	143	314	75
23 weeks	6,113	5,098	5,897	64,459	333,967	165,405	79,259	551,742	466,940	5,018	11,197	1,791

[1] New York City only.
[2] Period ended earlier than Saturday.

Telegraphic morbidity reports from State health officers for the week ended June 9, 1945, and comparison with corresponding week of 1944 and 5-year median—Con.

Division and State	Poliomyelitis			Scarlet fever			Smallpox			Typhoid and paratyphoid fever [2]		
	Week ended—		Median 1940–44	Week ended—		Median 1940–44	Week ended—		Median 1940–44	Week ended—		Median 1940–44
	June 9, 1945	June 10, 1944		June 9, 1945	June 10, 1944		June 9, 1945	June 10, 1944		June 9, 1945	June 10, 1944	
NEW ENGLAND												
Maine	0	0	0	38	40	3	0	0	0	0	0	0
New Hampshire	1	0	0	7	3	3	0	0	0	0	0	0
Vermont	0	0	0	10	2	5	0	0	0	0	0	0
Massachusetts	1	0	0	312	251	197	0	0	0	5	3	4
Rhode Island	0	0	0	5	8	8	0	0	0	0	0	0
Connecticut	0	0	1	45	43	39	0	0	0	1	1	1
MIDDLE ATLANTIC												
New York	11	4	3	526	304	344	0	0	0	7	6	7
New Jersey	0	0	0	112	141	141	0	0	0	1	0	2
Pennsylvania	1	0	0	412	219	219	0	0	0	4	6	6
EAST NORTH CENTRAL												
Ohio	0	2	0	336	274	229	1	0	1	4	3	4
Indiana	1	0	0	[4] 64	59	42	[4] 0	0	1	1	0	0
Illinois	2	2	2	205	146	146	0	0	1	1	6	3
Michigan [3]	0	0	0	234	215	178	0	0	0	1	5	3
Wisconsin	0	0	0	176	151	93	0	1	1	0	1	1
WEST NORTH CENTRAL												
Minnesota	0	0	0	77	61	40	0	0	0	0	2	0
Iowa	0	0	0	28	127	14	0	0	0	0	1	0
Missouri	0	0	0	44	29	37	0	0	0	0	0	2
North Dakota	0	0	0	18	14	4	0	1	0	1	0	0
South Dakota	0	0	0	26	7	8	1	0	0	0	0	0
Nebraska	0	0	0	28	19	13	0	0	0	0	0	0
Kansas	0	0	0	43	34	27	0	0	0	0	1	1
SOUTH ATLANTIC												
Delaware	0	0	0	3	4	4	0	0	0	0	0	0
Maryland [3]	0	0	0	125	83	36	0	0	0	1	1	1
District of Columbia	1	0	0	21	32	8	0	0	0	0	1	0
Virginia	2	0	0	65	32	20	0	0	0	0	3	5
West Virginia	1	0	0	36	51	18	0	0	0	2	2	3
North Carolina	2	1	0	41	15	15	0	0	0	3	0	0
South Carolina	3	0	0	12	4	4	0	0	0	1	1	1
Georgia	0	1	0	14	9	9	0	0	0	0	2	9
Florida	1	1	1	2	3	1	0	0	0	5	2	4
EAST SOUTH CENTRAL												
Kentucky	0	5	1	25	23	35	0	1	0	5	5	4
Tennessee	2	1	1	31	28	28	0	0	0	1	6	4
Alabama	2	0	1	13	8	8	0	0	0	9	1	1
Mississippi [3]	0	3	0	5	6	3	0	0	0	0	0	2
WEST SOUTH CENTRAL												
Arkansas	1	0	0	3	2	2	2	0	1	0	5	5
Louisiana	1	7	1	19	2	4	1	0	0	4	4	4
Oklahoma	1	3	0	23	10	7	0	0	0	1	9	3
Texas	42	0	0	40	68	21	0	0	0	9	8	11
MOUNTAIN												
Montana	0	0	0	10	17	10	0	0	0	0	1	0
Idaho	0	0	0	7	25	7	0	0	0	0	2	0
Wyoming	0	0	0	4	25	5	0	0	0	0	0	0
Colorado	0	1	0	38	43	21	0	0	0	0	0	0
New Mexico	0	0	0	6	7	3	0	0	0	2	5	2
Arizona	0	0	0	8	24	6	0	0	0	2	2	1
Utah [3]	1	0	0	11	53	7	0	0	0	0	0	0
Nevada	0	0	0	0	0	0	0	0	0	0	0	0
PACIFIC												
Washington	2	1	1	47	129	20	0	0	0	0	1	0
Oregon	0	0	0	17	45	11	1	0	0	1	0	1
California	13	9	9	326	270	111	0	0	0	8	8	3
Total	92	41	41	3,698	3,165	2,338	6	3	9	80	104	116
23 weeks	903	586	586	[4] 120,416	135,274	87,636	[4] 224	251	553	1,401	1,790	1,939

[2] Period ended earlier than Saturday.
[3] Including paratyphoid fever reported separately as follows: Massachusetts 5; New York 2; Michigan 1; Florida 4; Kentucky 1; Texas 1; California 1.
[4] Corrections: Indiana, week ended May 5, scarlet fever 94 cases (instead of 89); smallpox 9 cases (instead of 14).

Telegraphic morbidity reports from State health officers for the week ended June 9, 1945, and comparison with corresponding week of 1944, and 5-year median—Con.

Division and State	Whooping cough			Week ended June 9, 1945							
	Week ended—			Dysentery			Encephalitis, infectious	Rocky Mt. spotted fever	Tularemia	Typhus fever	Undulant fever
	June 9, 1945	June 10, 1944	Median 1940–44	Amebic	Bacillary	Unspecified					
NEW ENGLAND											
Maine	41	1	14	0	0	0	0	0	0	0	0
New Hampshire	0	0	6	0	0	0	0	0	0	0	0
Vermont	32	7	7	0	0	0	0	0	0	0	1
Massachusetts	171	56	162	0	0	0	0	0	0	0	4
Rhode Island	16	11	27	0	0	0	1	0	0	0	1
Connecticut	41	53	53	0	0	0	0	0	0	0	1
MIDDLE ATLANTIC											
New York	210	155	279	5	21	0	0	0	0	0	6
New Jersey	112	61	122	1	0	0	0	0	0	0	0
Pennsylvania	166	66	237	0	0	0	0	1	0	0	4
EAST NORTH CENTRAL											
Ohio	130	72	145	1	1	0	0	0	0	1	1
Indiana	34	16	35	0	0	0	0	0	0	0	1
Illinois	48	34	102	3	8	0	2	3	0	0	12
Michigan [2]	45	81	218	0	0	0	0	0	0	0	7
Wisconsin	26	48	125	0	0	0	0	0	0	0	6
WEST NORTH CENTRAL											
Minnesota	11	22	29	2	0	0	0	0	0	0	3
Iowa	0	4	23	0	0	0	0	0	0	0	0
Missouri	29	19	20	0	0	1	0	0	1	0	0
North Dakota	0	3	11	0	0	0	1	0	0	0	0
South Dakota	0	6	4	0	0	0	0	0	0	0	3
Nebraska	0	2	11	0	0	0	0	0	0	0	0
Kansas	31	30	55	0	0	0	0	0	1	0	8
SOUTH ATLANTIC											
Delaware	1	0	1	0	0	0	0	0	0	0	0
Maryland [2]	88	45	108	0	0	1	0	1	0	0	1
District of Columbia	3	1	11	0	0	0	0	0	0	0	0
Virginia	132	35	59	0	0	60	0	3	3	1	1
West Virginia	11	23	58	0	0	0	0	1	0	0	0
North Carolina	158	124	160	0	4	0	1	0	0	1	1
South Carolina	75	79	79	3	27	0	0	1	1	2	0
Georgia	21	30	27	1	6	1	0	0	1	14	5
Florida	8	19	10	4	0	0	0	0	0	6	0
EAST SOUTH CENTRAL											
Kentucky	23	76	76	0	0	0	0	1	0	0	0
Tennessee	33	21	51	0	0	2	0	2	0	0	0
Alabama	67	14	39	0	0	0	0	0	0	18	4
Mississippi [2]	----	----	----	0	0	0	0	0	0	0	1
WEST SOUTH CENTRAL											
Arkansas	18	26	42	0	1	0	0	0	0	0	0
Louisiana	5	3	5	3	0	0	0	0	1	1	4
Oklahoma	9	4	12	0	0	0	1	0	0	0	1
Texas	266	230	294	13	487	77	0	0	0	53	16
MOUNTAIN											
Montana	3	4	6	0	0	0	0	0	0	0	0
Idaho	1	9	8	0	0	0	0	0	0	0	0
Wyoming	2	9	7	0	0	0	0	0	2	0	0
Colorado	40	13	25	0	0	0	0	2	0	0	1
New Mexico	6	7	20	0	0	0	0	0	0	0	0
Arizona	11	8	23	0	0	24	0	0	0	0	0
Utah [2]	25	84	65	0	0	0	0	0	2	0	1
Nevada	0	0	2	0	0	6	0	0	0	0	0
PACIFIC											
Washington	17	15	65	0	0	0	0	0	0	0	0
Oregon	24	14	20	0	0	0	0	0	0	0	1
California	489	96	431	4	1	0	1	0	0	0	0
Total	2,679	1,736	3,778	40	556	172	7	15	12	97	95
Same week 1944	1,736	----	----	29	738	266	15	18	19	102	82
Average 1942–44	3,251	----	----	50	410	172	14	[3] 22	22	[3] 43	----
23 weeks: 1945	57,437	----	----	711	9,918	2,690	156	90	363	1,270	2,062
1944	41,503	----	----	591	6,740	1,909	256	89	257	1,101	1,415
Average 1942–44	74,281	----	[3] 88,081	589	4,555	1,442	238	[3] 127	366	[3] 826	----

[2] Period ended earlier than Saturday.
[3] 5-year median, 1940–44.

Anthrax: New York 1 case. *Leprosy:* California 1 case.

WEEKLY REPORTS FROM CITIES

City reports for week ended June 2, 1945

This table lists the reports from 89 cities of more than 10,000 population distributed throughout the United States, and represents a cross section of the current urban incidence of the diseases included in the table.

	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
NEW ENGLAND												
Maine:												
Portland	0	0	------	0	0	0	2	0	4	0	0	11
New Hampshire:												
Concord	0	0	------	0	1	0	0	0	0	0	0	0
Vermont:												
Barre	0	0	------	0	13	0	0	0	0	0	0	0
Massachusetts:												
Boston	1	0	------	0	93	2	12	0	69	0	0	31
Fall River	0	0	------	0	0	1	0	0	3	0	0	3
Springfield	0	0	------	0	1	0	2	0	14	0	0	1
Worcester	0	0	------	0	8	0	10	0	2	0	0	3
Rhode Island:												
Providence	0	0	1	0	9	1	2	0	5	0	0	22
Connecticut:												
Bridgeport	0	0	------	0	0	0	0	0	7	0	0	0
Hartford	0	0	------	0	31	0	0	0	15	0	0	4
New Haven	0	0	------	0	0	0	1	0	0	0	0	11
MIDDLE ATLANTIC												
New York:												
Buffalo	0	0	------	0	6	0	4	0	5	0	0	5
New York	10	0	4	0	39	6	62	2	247	0	1	66
Rochester	0	0	------	1	41	0	2	0	18	0	0	12
Syracuse	0	0	------	0	0	2	3	0	8	0	0	12
New Jersey:												
Camden	1	0	------	0	2	0	0	0	1	0	0	1
Newark	0	0	1	0	3	0	5	0	18	0	0	7
Trenton	0	0	------	0	13	0	5	0	2	0	0	0
Pennsylvania:												
Philadelphia	1	0	------	0	414	4	14	0	83	0	0	76
Pittsburgh	0	0	1	0	0	1	6	0	52	0	0	10
Reading	0	0	1	0	2	0	0	0	15	0	0	0
EAST NORTH CENTRAL												
Ohio:												
Cincinnati	0	0	------	1	4	5	11	0	11	0	0	10
Cleveland	0	0	1	0	4	1	10	0	45	0	0	17
Columbus	0	0	------	0	2	2	3	0	3	0	0	8
Indiana:												
Fort Wayne	0	0	------	0	0	0	2	0	7	0	0	0
Indianapolis	4	0	------	2	14	0	4	0	6	0	0	1
South Bend	0	0	------	0	1	0	0	0	3	0	0	0
Terre Haute	0	0	------	0	0	1	0	0	3	0	0	0
Illinois:												
Chicago	1	0	------	0	221	10	28	0	104	0	0	26
Springfield	0	0	------	0	1	0	3	0	2	0	0	0
Michigan:												
Detroit	9	1	1	0	98	1	14	0	114	0	0	7
Flint	0	0	------	0	2	0	4	0	6	0	0	0
Grand Rapids	0	0	------	0	3	0	0	0	5	0	0	1
Wisconsin:												
Kenosha	0	0	------	0	21	0	0	0	2	0	0	2
Milwaukee	0	0	------	0	38	0	5	0	76	0	0	1
Racine	0	0	------	0	2	0	0	0	2	0	0	1
Superior	0	0	------	0	16	0	0	0	3	0	0	2
WEST NORTH CENTRAL												
Minnesota:												
Duluth	2	0	------	0	0	0	0	0	11	0	0	0
Minneapolis	0	0	------	1	15	0	6	0	17	0	0	3
St. Paul	0	0	------	0	1	0	2	0	5	0	0	0
Missouri:												
Kansas City	2	0	------	0	8	1	5	0	7	0	0	0
St. Joseph	0	0	------	0	2	0	0	0	1	0	3	0
St. Louis	3	0	------	3	7	5	11	0	16	0	0	8

City reports for week ended June 2, 1945—Continued

	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
WEST NORTH CENTRAL—continued												
North Dakota:												
Fargo	0	0	------	0	0	0	0	0	2	0	0	1
Nebraska:												
Omaha	0	0	------	0	13	1	3	0	15	0	0	0
Kansas:												
Topeka	0	0	------	0	3	0	2	0	5	0	0	1
Wichita	0	0	------	0	0	0	3	0	8	0	0	0
SOUTH ATLANTIC												
Delaware:												
Wilmington	1	0	------	0	1	2	1	0	1	0	0	0
Maryland:												
Baltimore	4	0	------	0	13	0	11	0	60	0	1	61
Cumberland	0	0	------	0	1	0	0	0	2	0	0	0
Frederick	0	0	------	0	0	0	0	0	0	0	0	0
District of Columbia:												
Washington	0	0	------	1	6	3	4	0	25	0	0	11
Virginia:												
Lynchburg	0	0	------	0	0	0	0	0	0	0	1	0
Richmond	0	0	------	0	3	0	2	1	12	0	0	0
Roanoke	0	0	------	0	1	0	0	0	2	0	0	0
West Virginia:												
Charleston	0	0	------	0	0	0	0	0	1	0	0	0
Wheeling	0	0	------	0	2	0	0	0	0	0	0	0
North Carolina:												
Raleigh	0	0	------	0	4	0	4	0	1	0	0	5
Wilmington	0	0	------	0	2	0	0	0	1	0	0	5
Winston-Salem	0	0	------	0	0	0	1	0	8	0	0	5
South Carolina:												
Charleston	0	0	1	0	0	1	0	0	0	0	0	0
Georgia:												
Atlanta	0	0	1	1	0	1	5	0	4	0	0	5
Brunswick	0	0	------	0	0	0	0	0	0	0	0	2
Savannah	0	0	------	0	0	0	0	1	2	0	1	0
Florida:												
Tampa	0	0	------	1	1	0	3	0	0	0	0	0
EAST SOUTH CENTRAL												
Tennessee:												
Memphis	0	0	2	0	36	3	3	0	4	0	0	16
Nashville	0	0	------	0	0	0	0	0	3	0	0	0
Alabama:												
Birmingham	0	0	------	0	0	1	0	1	1	0	1	0
Mobile	0	0	------	0	0	0	2	0	0	0	0	0
WEST SOUTH CENTRAL												
Arkansas:												
Little Rock	1	0	------	0	5	0	0	0	1	0	0	0
Louisiana:												
New Orleans	0	0	1	1	7	1	4	0	3	0	1	2
Shreveport	0	0	------	0	0	1	6	0	1	0	0	0
Texas:												
Dallas	2	0	------	0	6	0	0	1	2	0	0	12
Houston	3	0	------	0	0	0	3	3	3	0	2	2
San Antonio	1	0	------	0	0	1	4	0	2	0	0	1
MOUNTAIN												
Montana:												
Billings	0	0	------	0	3	0	0	0	6	0	0	0
Great Falls	0	0	------	0	1	0	0	0	2	0	0	0
Helena	0	0	------	0	0	0	0	0	0	0	0	0
Missoula	0	0	------	0	1	0	0	0	1	0	0	0
Idaho:												
Boise	0	0	------	0	0	0	0	0	0	0	0	0
Colorado:												
Denver	0	0	4	0	3	0	2	0	19	0	0	17
Pueblo	0	0	------	0	0	0	2	0	0	0	0	0
Utah:												
Salt Lake City	0	0	------	0	115	0	2	0	4	0	0	7

City reports for week ended June 2, 1945—Continued

	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
PACIFIC												
Washington:												
Seattle	1	0	------	0	25	0	3	0	9	0	0	1
Spokane	0	0	------	0	4	0	0	0	3	0	0	0
Tacoma	0	0	------	0	24	0	1	0	2	0	0	0
California:												
Los Angeles	2	0	2	1	59	2	3	0	49	0	0	22
Sacramento	0	0	1	1	9	0	0	0	12	0	0	2
San Francisco	0	0	1	0	177	0	3	0	27	0	1	14
Total	49	1	23	14	1,661	60	315	9	1,320	0	12	554
Corresponding week, 1944	45	------	18	11	3,259	------	258	------	1,117	1	18	294
Average, 1940–44	59	------	43	[1] 14	[2] 4,908	------	[1] 294	------	1,178	1	20	1,050

[1] 3-year average, 1942–44.
[2] 5-year median, 1940–44.

Anthrax.—Cases: Philadelphia, 1.
Dysentery, amebic.—Cases: New York, 3; Atlanta, 1; Los Angeles, 1.
Dysentery, bacillary.—Cases: New York, 1.
Dysentery, unspecified.—Cases: San Antonio, 17.
Rocky Mountain spotted fever.—Cases: Baltimore, 1; Denver, 1.
Tularemia.—Cases: St. Louis, 1.
Typhus fever, endemic.—Cases: Savannah, 2; Tampa, 1; New Orleans, 3; Houston, 1; San Antonio, 3.

Rates (annual basis) per 100,000 population, by geographic groups, for the 89 cities in the preceding table (estimated population, 1943, 34,324,200)

	Diphtheria case rates	Encephalitis, infectious, case rates	Influenza		Measles case rates	Meningitis, meningococcus, case rates	Pneumonia death rates	Poliomyelitis case rates	Scarlet fever case rates	Smallpox case rates	Typhoid and paratyphoid fever case rates	Whooping cough case rates
			Case rates	Death rates								
New England	2.6	0.0	2.6	0.0	408	10.5	75.8	0.0	311	0.0	0.0	225
Middle Atlantic	5.6	0.0	3.2	0.5	241	6.0	46.7	0.9	208	0.0	0.5	87
East North Central	8.5	0.6	1.2	1.8	260	12.2	51.1	0.0	238	0.0	0.0	46
West North Central	13.9	0.0	0.0	8.0	97	13.9	63.7	0.0	173	0.0	6.0	26
South Atlantic	8.2	0.0	3.3	4.9	56	11.4	50.7	3.3	195	0.0	4.9	154
East South Central	0.0	0.0	11.8	0.0	212	23.6	29.5	5.9	47	0.0	5.9	94
West South Central	21.0	0.0	3.0	3.0	54	9.0	50.7	11.9	36	0.0	9.0	51
Mountain	0.0	0.0	31.8	0.0	977	0.0	47.7	0.0	254	0.0	0.0	191
Pacific	4.7	0.0	6.3	3.2	471	3.2	15.8	0.0	161	0.0	1.6	62
Total	7.5	0.2	3.5	2.1	253	9.1	48.0	1.4	201	0.0	1.8	84

FOREIGN REPORTS

CANADA

Provinces—Communicable diseases—Week ended May 19, 1945.—During the week ended May 19, 1945, cases of certain communicable diseases were reported by the Dominion Bureau of Statistics of Canada as follows:

Disease	Prince Edward Island	Nova Scotia	New Brunswick	Quebec	Ontario	Manitoba	Saskatchewan	Alberta	British Columbia	Total
Chickenpox		27		214	348	56	28	56	77	806
Diphtheria		2	2	37	1	4		1	3	50
Dysentery:										
Bacillary				5						5
Unspecified					16					16
Encephalitis, infectious						1				1
German measles				3	36	2	4	26	56	127
Influenza		18			62	4			19	103
Measles		5	1	119	177	26	71	28	350	777
Meningitis, meningococcus		2	1	2					2	7
Mumps		5		224	110	42	16	119	11	527
Poliomyelitis				1						1
Scarlet fever		8	33	70	102	11	1	28	27	280
Tuberculosis (all forms)		4	4	164	53	22	25	12		284
Typhoid and paratyphoid fever				8				1		9
Undulant fever				3	2				1	6
Venereal diseases:										
Gonorrhea	1	40	46	140	135	33	34	28	75	532
Syphilis		28	6	139	87	4	7	12	61	344
Whooping cough		1		110	15	1	2	14	6	149

CUBA

Habana—Communicable diseases—4 weeks ended May 26, 1945.—During the 4 weeks ended May 26, 1945, certain communicable diseases were reported in Habana, Cuba, as follows:

Disease	Cases	Deaths	Disease	Cases	Deaths
Chickenpox	16		Tuberculosis	1	
Diphtheria	14		Typhoid fever	61	5
Measles	2				

SWEDEN

Notifiable diseases—March 1945.—During the month of March 1945, cases of certain notifiable diseases were reported in Sweden as follows:

Disease	Cases	Disease	Cases
Cerebrospinal meningitis	6	Poliomyelitis	49
Diphtheria	233	Scarlet fever	1,643
Dysentery, epidemic	36	Syphilis	138
Gonorrhea	1,057	Undulant fever	4
Hepatitis, epidemic	747	Weil's disease	4
Paratyphoid fever	4		

WORLD DISTRIBUTION OF CHOLERA, PLAGUE, SMALLPOX, TYPHUS FEVER, AND YELLOW FEVER

From medical officers of the Public Health Service, American consuls, International Office of Public Health, Pan American Sanitary Bureau, health section of the League of Nations, and other sources. The reports contained in the following tables must not be considered as complete or final as regards either the list of countries included or the figures for the particular countries for which reports are given.

CHOLERA

[C indicates cases; P, present]

NOTE.—Since many of the figures in the following tables are from weekly reports, the accumulated totals are for approximate dates.

Place		January–March 1945	April 1945	May 1945—week ended—			
				5	12	19	26
ASIA							
India	C	24,022					
Bombay	C	8	6	4	4	9	
Calcutta	C	773	1,760	302	232	241	232
Cawnpore	C	1	5	5	13		
Chittagong	C	8	5				
Delhi	C	[1] 1		2			
Madras	C	45	2				
Vizagapatam	C				13		
Indochina: Cochinchina	C				P		

[1] Imported.

PLAGUE

[C indicates cases]

AFRICA							
Algeria	C	[1] 12					
Basutoland	C	4					
Bechuanaland	C	7					
Belgian Congo	C	4		1			
British East Africa:							
Kenya	C	3			1		
Uganda	C	2	2				
Egypt	C	13	21	2	7	16	
Port Said	C	5	1		4	5	
Suez	C	2	1	2	2	1	
French West Africa	C	5					
Dakar	C	1					
Madagascar	C	63	39				
Morocco (French)	C	107	14			[2] 32	
Senegal	C	54					
Tunisia	C	3					
Union of South Africa	C	5	1				
ASIA							
India	C	15,810					
Iraq	C	34					
Palestine	C	9	1				
Plague-infected rats		16					
EUROPE							
France: Corsica—Ajaccio	C					2	
Portugal: Azores	C	3					
Spain: Canary Islands	C	1					
SOUTH AMERICA							
Bolivia: Santa Cruz Department	C	[3] 10	4			[4] 61	
Ecuador:							
Chimborazo Province	C	3	3				
Loja Province	C		2				
Peru:							
Ancash Department	C	1					
Ica Department	C	[5] 1					
Lambayeque Department	C	12					
Libertad Department	C	8					
Lima Department	C	8					
Piura Department	C	3					
OCEANIA							
Hawaii Territory	C		1				
Plague-infected rats [6]	C	8	1				

[1] Includes 1 case of pneumonic plague.
[2] For the period May 1–20, 1945.
[3] Includes 4 confirmed cases.
[4] Date not specified.
[5] Suspected.
[6] Plague infection was also proved positive in a pool of 5 mice on Jan. 4, in a pool of fleas on Feb. 14, and in a pool of 40 fleas on Mar. 14, 1945.

SMALLPOX

[C indicates cases; P, present]

Place		January–March 1945	April 1945	May 1945—week ended—			
				5	12	19	26
AFRICA							
Algeria	C	94	16				
Angola	C	54					
Basutoland	C	79	164				
Belgian Congo	C	2,279	342				
British East Africa:							
Kenya	C	96	20	4			
Nyasaland	C	3	6				
Tanganyika	C	1,034	1,337	139			
Uganda	C	363	103	36			
Cameroon (French)	C	251	40				
Dahomey	C	47	45				
Egypt	C	506	128		49		
French Equatorial Africa	C	1,253	219				
French Guinea	C	769	233				
French West Africa	C	248	71				
Gambia	C	26	30	2	9		
Gold Coast	C	26					
Ivory Coast	C	133	107				
Mauritania	C	2	39				
Morocco (French)	C	118	61				
Nigeria	C	1,970	198				
Niger Territory	C	202	108				
Rhodesia, Northern	C	349	332		2		
Senegal	C	181	130				
Sudan (Anglo-Egyptian)	C	[1] 3					
Sudan (French)	C	683	356				
Togo (British)	C	25					
Togo (French)	C	308	46				
Union of South Africa	C	395	P				
ASIA							
Arabia	C	16				3	
Ceylon	C	[2] 320	21	7			
China: Kunming (Yunnan Fu)	C	5	1			1	
India	C	112,990					
Iran	C	296					
Iraq	C	11	2	2			6
Syria and Lebanon	C	6					
EUROPE							
Belgium	C	1					
France	C	2					
Great Britain: Scotland	C	[1] 1	1				
Italy	C	809	85	13			
Portugal	C	2	8		5	1	
Spain	C	23	1				
Turkey	C	259	15		2	1	5
NORTH AMERICA							
Canada	C	6					
Guatemala	C	1	2				
Honduras	C		1				
Mexico	C	483					
Nicaragua	C	123					
SOUTH AMERICA							
Bolivia	C	97					
Brazil	C	45	6				
Colombia	C	67	46	13	15	9	
Ecuador	C	11	2		5	3	
Paraguay	C	1					
Peru	C	12					
Venezuela	C	[3] 367	[3] 69				

[1] Imported.
[2] Includes some cases of chickenpox.
[3] Reported as alastrim.

TYPHUS FEVER*

[C indicates cases; P, present]

Place		January–March 1945	April 1945	May 1945—week ended— 5	12	19	26
AFRICA							
Algeria	C	620	86				
Basutoland	C	41	7				
Belgian Congo [1]	C	19	3				
British East Africa: Kenya	C	16					
Egypt	C	6,510	2,028		934		
French West Africa: Dakar [1]	C	2	1				
Libya: Tripolitania	C	11	6				
Morocco (French)	C	2,185	1,112			[3] 603	
Rhodesia, Northern	C	11					
Tunisia	C	88	272				
Union of South Africa	C	158	P				
ASIA							
China: Kunming (Yunnan Fu)	C	28	4				
India	C	17					
Iran	C	255	30				
Iraq	C	44	33	23	32		18
Palestine [1]	C	20	3	1			
Syria and Lebanon	C	7	1				
Trans-Jordan	C	26	7				
EUROPE							
Albania	C	[2] 100					
Belgium	C		1				
Bulgaria	C	477	293	27			
France	C	5					
Gibraltar	C	4					
Great Britain	C	[4] 1			1		
Greece	C	23	3				
Italy	C	13	1			35	
Malta and Gozo [1]	C	6					
Netherlands	C		6				
Portugal	C	35	4	1		1	
Rumania	C	[5] 7,831					
Slovakia	C	230					
Spain	C	6	2				
Turkey	C	1,239	244	71	89	79	61
Yugoslavia	C	137					
NORTH AMERICA							
Canada [1]	C	1					
Costa Rica	C	2					1
Cuba [1]	C	1				1	
Guatemala	C	487	172				
Jamaica	C	9	3	2			
Mexico	C	494					
Panama (Republic)	C	1					
Puerto Rico [1]	C	14	6				
Virgin Islands [1]	C	4					
SOUTH AMERICA							
Bolivia	C	88					
Brazil	C	1					
Chile [1]	C	135					
Colombia	C	18		2			
Curacao	C	1					
Ecuador	C	109	51				
Peru	C	131					
Venezuela [1]	C	35	15				
OCEANIA				—			
Australia	C	70	9				
Hawaii Territory	C	23	1		2		

*Reports from some areas are probably murine type, while others probably include both murine and louse-borne types.

[1] Reports cases as murine type.
[2] For the period May 1–20, 1945.
[3] For the months of February and March 1945.
[4] Imported.
[5] For the period Jan. 1–20, 1945.

YELLOW FEVER

[C indicates cases; D, deaths]

Place		January-March 1945	April 1945	May 1945—week ended—			
				5	12	19	26
AFRICA							
Ivory Coast:							
Gaoua.[1]							
Guiglo	C	1					
SOUTH AMERICA							
Brazil:							
Goiaz State	D	73	2				
Minas Geraes State	D	10	7				
Colombia: Santander del Norte Department	D	[2] 5					
Peru: Cuzco Department	C	1	1				
Venezuela:							
Bolivar State	C		1				
Tachira State	D	2					

[1] For the week ended June 2, 1945, 1 fatal case of suspected yellow fever was reported in Gaoua, Ivory Coast.
[2] For the period Jan. 1 to Mar. 11, 1945.

FEDERAL SECURITY AGENCY

UNITED STATES PUBLIC HEALTH SERVICE

THOMAS PARRAN, *Surgeon General*

DIVISION OF PUBLIC HEALTH METHODS

G. ST. J. PERROTT, *Chief of Division*

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